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TITLE: GROWTH DYNAMICS OF BREAST CANCER CELLS: A STUDY OF  
GROWTH REGULATORY FACTORS

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The contract for purchase of a flow cytometer/cell sorter was awarded to Becton-Dickinson. This instrument was installed and made operational during the first week in September 1993. The contract for purchase of an image analysis system was awarded to Roche Image Analysis Systems and the instrument was installed and made operational during the last week in August 1993. A microtome/cryostat was purchased from Zeiss Instruments Co. and was made operational in mid-September 1993.

**Scientific Progress:** Due to a prolonged contracting process in the purchase of our image analysis system and to an even longer and more involved contracting process that required extensive and technically demanding justification in the purchase of our flow cytometer/cell sorter, we have been delayed almost 1 year in initiating our studies of breast cancer cells.

At the present time, we have our flow cytometer/cell sorter in place and operational and we have our image analysis system in place and operational. Ms. Rebecca Smiley, GS-9 Microbiologist, received formal training in the operation of our flow cytometer/cell sorter at the Becton-Dickinson facility in Boston, Massachusetts in mid-September 1993. Dr. Veit, PI, and Ms. McIntyre, GS-7 Medical Technologist, received formal training in the operation of our image analysis system at the Roche Image Analysis Systems facility in Research Triangle, North Carolina during the first week in October 1993.

Several human breast cancer cell lines have been obtained from the American Type Culture Collection Company and are currently being maintained by serial passage in *in vitro* culture in our laboratory. Samples of all lines have been frozen in 10% DMSO and are being stored in our liquid nitrogen repository. These cell lines have been utilized for establishing the various staining procedures for the immunohistochemical (image analysis) and flow cytometric analyses of DNA (S-phase fraction, cell cycle, and ploidy), epidermal growth factor receptor expression and production, estrogen/progesterone receptor expression, HER-2/neu expression, Ki-67 expression, PCNA expression, transforming growth factor expression, and insulin-like growth factor expression.

**Future Plans:** Additional training may be required for the implementation of our DNA modeling software (Modfit) in our flow cytometric studies of DNA. Due to the delay in onset of our studies because of a prolonged contracting process in obtaining our flow cytometer/cell sorter and image analysis system, the PI will request that a no-cost extension of 18 months be granted by the contracting officer for this project (that the termination of this contract be extended from September 30, 1994 to March 31, 1996). The personnel involved in this project are fully trained (with the exception of DNA modeling training) in the operation of our flow cytometer/cell sorter and image analysis system and are currently involved in the analysis of established breast cancer cell lines and primary breast cancer biopsies with respect to growth factor requirements and/or growth factor receptor expression. *In vivo* studies, utilizing growth of breast tumors in nude mice, as well as studies of apoptosis, will be initiated within the next six months.